

## RED DEER ANTLERS AS A SOURCE OF REGENERATIVE CELLS

УДК 612.014

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## ПАНТЫ МАРАЛА КАК ИСТОЧНИК ПОЛУЧЕНИЯ РЕГЕНЕРАТИВНЫХ КЛЕТОК

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The first reference on the use of antlers for medical purposes was found in a Khan's tomb, dating back to 168 AD, in Huang Province (China). The silk scroll contained 52 prescriptions for potions using antlers and venison (1). In the 60s of the twentieth century, 300 preparations were already created based on the products of reindeer husbandry (2, 3). Antlers were used to treat ulcers, burns, carbuncles, epilepsy, to increase vitality, endurance, strengthen teeth (4).

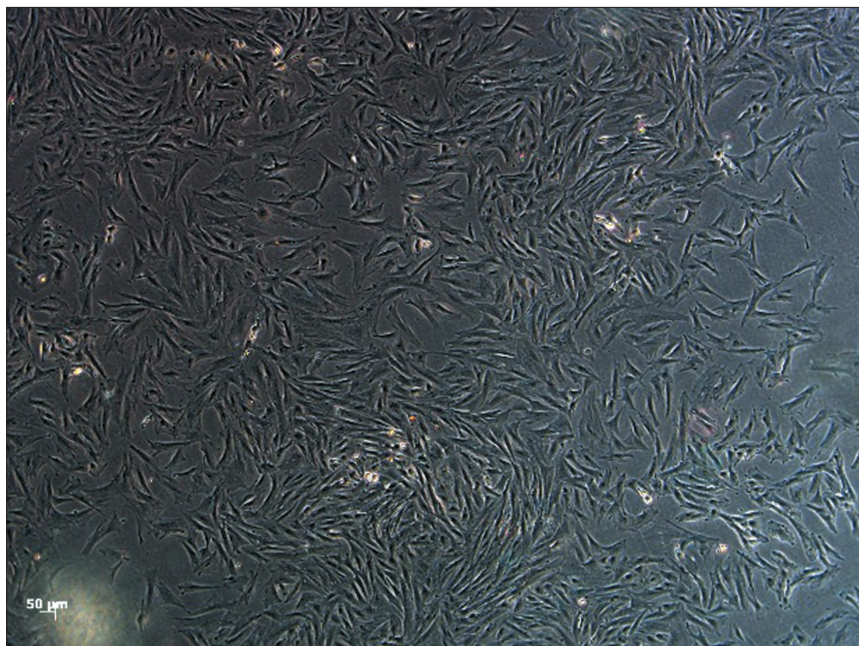
High prices and demand for antler products on the international market led to a massive extermination of the red deers of Russia (5). By the end of the 19th century, over 150 thousand red deers were killed for the antlers in Siberia and the Far East. (1) For the further successful cropping of antlers, the domestication of animals was necessary. By 2005, there were 184 farms specialized in antler-reindeer farming in Russia, receiving over 45 tons of canned antlers annually, with 70% of all products produced in Altai (6). According to the Republican target program "Development of antler rein-

deer husbandry in the Altai Republic 2011–2020" of August 8, 2011, the expected indicators by 2020 should be an increase in the number of deer to 60 thousand heads and an increase in the production of raw antlers to 145 tons.

The active development of regenerative medicine and cellular technologies determines the search for new sources of regenerative cells and maral antlers can become such source (7).

### Materials and methods

The biomaterial was collected (red deer antlers) under local anesthesia. The resulting biomaterial was divided into 1 × 1 cm sterile scalpel and transferred to sterile transport medium tubes (DMEM supplemented with 2% FBS, 2 mM L-glutamine, 200 U / ml penicillin, and 200 µg / ml streptomycin, 200 units / ml amphotericin, 100 units / ml gentamicin). The material was delivered to the laboratory within 10 hours after collection. In the laboratory, under sterile conditions,



**Fig. 1.** Cell line of regenerative cells of red deer antlers. Light microscopy. × 50

antlers were finely dispersed and subjected to enzymatic treatment in 0.15% type II collagenase. Next, the cell suspension was washed from the enzyme and the viability assessment was performed. After the calculations, the cells were transferred to the culture flask, at the rate of  $3 \times 10^5$  cells /  $\text{cm}^2$ , and cultured in DMEM with 10% FBS, 2 mM L-glutamine, 100 units / ml penicillin and 100  $\mu\text{g}$  / ml streptomycin. The expansion of the cells was carried out according to the standard method of cultivation of human MMSC.

### Results

During the study, primary cell culture was obtained from maral antlers. The morphology of the cell line was characterized by homogeneous cells having a fibroblast-like shape (Fig. 1). (Place figure 1). The results of immunophenotyping showed that the cell culture was characterized by the absence of CD34, CD45 (hematopoietic) and the presence of CD105, CD90 (mesenchymal) surface markers. The average doubling time of the cell population was  $29 \pm 1$  hours. For comparison, this indicator for the culture of human fibroblast cells is  $42 \pm 3$  hours on a similar culture

medium. During the analysis of the effectiveness of colony formation (IVF), it was noted that the regenerative cells of the maral antlers form colonies that are morphologically similar to the colonies of human fibroblasts. IVF was  $42 \pm 8\%$ , versus  $34 \pm 3\%$  in fibroblasts. The study of the cytokine profile showed that maral antler cells have the same qualitative composition as human fibroblast-like cells. The difference was noted in the expression of vascular endothelial growth factor (VEGF). In human fibroblasts, this indicator is  $626.6 \pm 92.3$  pg / ml, while in the regenerative cells of maral antlers  $2467.7 \pm 114.8$  pg / ml.

### Conclusions

The results of the study showed that regenerative cells isolated from red deer antlers are of mesenchymal origin and, therefore, may be called mesenchymal stromal cells (MSCs). The regenerative potential of MSC antlers was higher than that of human skin fibroblasts. Thus, the use of this type of cells can be considered as a promising way to obtain a pharmacological substance or biologically active complexes for regenerative medicine.

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